

## Claims

1. A method of determining whether or not the binding of a molecule capable of binding to a ligand to the ligand is  
5 specific, which comprises the steps shown below:
  - (1) a step of treating a sample with a ligand-immobilized solid phase carrier to obtain a treated liquid, and extracting the protein bound onto the solid phase carrier to obtain a ligand-immobilized solid phase carrier extract 1,
  - 10 (2) a step of treating the treated liquid obtained in the previous step with a ligand-immobilized solid phase carrier (another solid phase carrier wherein the same kind of ligand as the ligand-immobilized solid phase carrier used in the previous step is immobilized) to obtain a treated liquid, and  
15 extracting the protein bound onto the solid phase carrier to obtain a ligand-immobilized solid phase carrier extract 2,
  - (3) a step of comparing and analyzing the proteins contained in the ligand-immobilized solid phase carrier extract 1 and the proteins contained in the ligand-immobilized solid phase  
20 carrier extract 2,
  - (4) a step of identifying a protein that is detected in the ligand-immobilized solid phase carrier extract 1, and that is not detected in the ligand-immobilized solid phase carrier extract 2 or, even if detected, shows a significantly greater  
25 reduction compared to other proteins than in the ligand-immobilized solid phase carrier extract 1, on the basis of the analytical results obtained in the step (3), and determining the protein to be specific for the ligand.
- 30 2. The method of claim 1, which comprises repeating the step (2) twice or more.
3. A method of determining whether or not the binding of a molecule capable of binding to a ligand to the ligand is

specific, which comprises the steps shown below:

- (1) a step of dividing a sample into two portions, and treating one thereof with an inert-substance-immobilized solid phase carrier to obtain a treated liquid,
- 5 (2) a step of treating the treated liquid after treatment with the inert-substance-immobilized solid phase carrier, obtained in the previous step, with a ligand-immobilized solid phase carrier (another solid phase carrier wherein the same kind of ligand as the ligand-immobilized solid phase carrier used in  
10 the step (3) and step (4) described below is immobilized) to obtain a treated liquid, and extracting the protein bound onto the solid phase carrier to obtain a ligand-immobilized solid phase carrier extract 1,
- (3) a step of treating the remaining portion of the sample  
15 divided into two portions in the step (1) with a ligand-immobilized solid phase carrier to obtain a treated liquid,
- (4) a step of treating the treated liquid after treatment with the ligand-immobilized solid phase carrier, obtained in the previous step, with a ligand-immobilized solid phase carrier  
20 (another solid phase carrier wherein the same kind of ligand as the ligand-immobilized solid phase carrier used in the previous step (3) is immobilized) to obtain a treated liquid, and extracting the protein bound onto the solid phase carrier to obtain a ligand-immobilized solid phase carrier extract 2,
- 25 (5) a step of comparing and analyzing the proteins contained in the ligand-immobilized solid phase carrier extract 1 and the proteins contained in the ligand-immobilized solid phase carrier extract 2,
- (6) a step of identifying a protein that is detected in the  
30 ligand-immobilized solid phase carrier extract 1, and that is not detected in the ligand-immobilized solid phase carrier extract 2 or, even if detected, shows a significantly greater reduction compared to other proteins than in the ligand-immobilized solid phase carrier extract 1, on the basis of the

analytical results obtained in the step (5), and determining the protein to be specific for the ligand.

4. The method of claim 3, wherein the inert substance is  
5 stearic acid.

5. The method of claim 3, wherein the inert substance is structurally similar to the subject ligand, and does not possess the physiological activity possessed by the ligand.

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6. The method of claim 1 or 3, wherein the sample is a biological sample.

7. The method of claim 1 or 3, which further comprises a step  
15 of calculating the binding constant of the protein in the sample to the ligand by comparison and analysis.